Multiple-sample microarray biochip

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Abstract

The multiple-sample microarray biochip has a solid carrier in the slide glass form and with separated and no-leakage cavities, which are formed by one layer of lattice plastic material adhered to thesurface of the slide glass and one layer of surface polyester film to seal the cavities. Each cavity has distributed microarray probes for simultaneously completing several reactions. The cavity has side length of multiple times of 3 mm and central interval of multiple times of 4.5 mm. The multiple-sample microarray biochip of the present invention is used for detection, analysis and identification in protein and polypeptide hybridization, nucleic acid hybridization and DNA cloning. Multiple sample and multiple item experiment may be performed parallelly.

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[54] 发明名称 多样品微阵列生物芯片

[57] 摘要

本发明涉及一种多样品微阵列生物芯片，其固态基底是一以载玻片形式的固相载体。载玻片的表面设有分隔设置的且不会互相渗漏的腔室，该腔室由一层格栅状的单面胶材粘贴于载玻片表面而形成，在格栅状单面胶材的表面再粘贴一层聚酯薄膜，以使腔室密闭。每个腔室内布有微阵列探针点，以同时完成多种反应。腔室的边长为 3mm 的倍数。两个相邻腔室的中心间距为 4.5mm 的倍数。本发明的多样品微阵列生物芯片用于蛋白质及多肽杂交、核酸杂交、脱氧核糖核酸扩增，能对生物分子进行并行检测、分析、鉴定及杂交反应，实现多样品多项目平行实验。

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1. 一种多样品微阵列生物芯片，其固态基底是一以载玻片形式的固相载体，其特征在于，所述载玻片的表面设有分隔设置的且不会互相渗漏的腔室，该腔室由一层格栅状的单面胶材粘贴于载玻片表面而形成，在格栅状单面胶材的表面再粘贴一层聚酯薄膜，以使上述腔室密闭；每个腔室内布有微阵列探针点，以同时完成多种反应。

2. 根据权利要求1所述的多样品微阵列生物芯片，其特征在于，所述多样品微阵列生物芯片用于蛋白质杂交、核酸杂交、脱氧核糖核酸扩增。

3. 根据权利要求1或2所述的多样品微阵列生物芯片，其特征在于，所述腔室的相邻两个腔室中心的间距为4.5mm。

4. 根据权利要求1或2所述的多样品微阵列生物芯片，其特征在于，所述腔室的相邻两个腔室中心的间距为9mm。

5. 根据权利要求1或2所述的多样品微阵列生物芯片，其特征在于，所述腔室的相邻两个腔室中心的间距为13.5mm。

6. 根据权利要求1或2所述的多样品微阵列生物芯片，其特征在于，所述腔室的相邻两个腔室中心的间距为18mm。

7. 根据权利要求1或2所述的多样品微阵列生物芯片，其特征在于，所述腔室设置为1×2个至4×10个。

多样品微阵列生物芯片

本发明涉及生命科学领域，特别涉及一种在固态基底上有序排列生物样品的多样品微阵列生物芯片。

现有的微阵列生物芯片，其固态支持物是未经特殊处理的平面介质，例如采用普通的载玻片或硝酸纤维素膜。这样的生物芯片，一个芯片一次反应中只能与一个非固态生物样品反应，当有许多样本需要进行相同的实验时，就要分别对每一个样品进行实验，如此不仅费时费力，而且处理条件(例如温度、反应时间等)的前后有细微差别，就会影响各样本之间的平行性和相互的可比性，因此无法达到多样品多项目平行实验的目的。利用这种生物芯片进行反应，非固态生物样本必须覆盖比较大的面积，因此需要较多的样本量，且反应过程中样品会流动、蒸发，由此会产生误差。

本发明的目的是提供一种多样品微阵列生物芯片，在该芯片上能对生物分子进行并行检测、分析、鉴定及杂交反应，实现多样品多项目平行实验。

本发明的技术方案如下：

一种多样品微阵列生物芯片，其固态基底是一以载玻片形式的固相载体，所述载玻片的表面设有分隔设置的且不会互相渗漏的腔室，该腔室由一层格栅状的单面胶材粘贴于载玻片表面而形成，在格栅状单面胶材的表面再粘贴一层聚酯薄膜，以使上述腔室密闭；每个腔室内布有微阵列探针点，以同时完成多种反应。

所述多样品微阵列生物芯片用于蛋白质杂交、脱氧核糖核酸杂交、脱氧核糖核酸扩增。

本发明所述的微阵列生物芯片是一种在固态基底即固态支持物上有序排列生物样品，对脱氧核糖核酸(DNA)、核糖核酸(RNA)、蛋白质或多肽等生物分子进行实验的器件，在该芯片上能对生物分子进行并行检测、分析、鉴定及杂交反应。产品体积小巧，便于储藏和使用，其制作和操作使用都很简便，生产成本低

廉，使用时可用通用的芯片扫描仪进行扫描。

本发明是一种新的反应模式，可用于蛋白质或 DNA 杂交、DNA 扩增等。由于在载玻片上设置了特殊的具有独立空间的小的腔室，在每个腔室中又布设了微阵列，点入不同的探针，如 DNA、RNA、蛋白质或多肽，因此可以达到多样品多项目平行实验的目的。如果用于临床检测分析，则可以进行多人份、多指标、多种疾病的同时诊断。

下面结合附图和实施例对本发明作详细说明。

图 1 是一种多样品微阵列生物芯片的主视结构示意图，腔室设置为 3×8 个。

图 2 是按图 1 所示的仰视结构示意图。

图 3 是粘贴在图 1 生物芯片上的聚酯薄膜结构示意图。

图 4 是一种设有 1×2 个腔室的多样品微阵列生物芯片示意图。

图 5 是一种设有 2×3 个腔室的多样品微阵列生物芯片示意图。

图 6 是一种设有 2×5 个腔室的多样品微阵列生物芯片示意图。

图 7 是一种设有 3×5 个腔室的多样品微阵列生物芯片示意图。

图 8 是一种设有 3×10 个腔室的多样品微阵列生物芯片示意图。

图 9 是一种设有 4×8 个腔室的多样品微阵列生物芯片示意图。

图 10 是在本发明的生物芯片上同时检测两个样品的示意图。

图 11 是图 10 中两个样品的放大图。

参看图 1 至图 3，本发明是一种多样品微阵列生物芯片，其固态基底是一以载玻片形式的固相载体。载玻片 1 是一种固体片状玻璃片，其尺寸与标准显微镜载玻片相同，为 25mm×75mm×0.96mm。

载玻片 1 的表面设有分隔设置的且不会互相渗漏的腔室 2，该腔室 2 由一层格栅状的单面胶材 3 粘贴于载玻片 1 表面而形成。在格栅状单面胶材 3 的表面再粘贴一层聚酯薄膜 4，以使上述腔室 2 密闭。在未贴置单面胶材 3 和聚酯薄膜 4 的载玻片 1 空白处可以粘贴标签，该空白处为载玻片 1 的贴标签处 5。

格栅状单面胶材 3 的大小为 50mm×25mm，以配合载玻片的尺寸。格栅状单面胶材 3 的厚度应至少 1mm，为 1mm～3mm，其中一面具有粘性，可与载玻片 1 表面紧密粘合，并将其表面分隔成不会互相渗漏的小的腔室 2。该单面胶材 3 浸泡在水中不会溶解，还可承受一定的水流冲击，因此粘贴于载玻片 1 后，能经受水流的清洗而不会变形、渗漏。在实验结束后，该单面胶材 3 将被揭除，以便使

用芯片扫描仪对芯片进行检测与分析。

聚酯薄膜 4 用于粘贴在格栅状单面胶材 3 的表面。聚酯薄膜 4 的一面具有粘性，可以粘贴在单面胶材 3 无粘性的一面，以达到将腔室 2 密闭的效果。聚酯薄膜 4 的粘性低于单面胶材 3，可以较容易地被撕除而不影响单面胶材 3 与载玻片 1 的粘贴。粘贴在载玻片 1 上的单面胶材 3 被覆盖粘贴聚酯薄膜 4 以后，形成了互相隔离的小的密闭腔室 2，腔室 2 之间毫无交叉污染，并降低了液体蒸发，可以在摇床上摇动进行充分反应，由此提高了反应的准确度。

每个腔室 2 的边长均为 3mm 的倍数。腔室 2 设置为 1×2 个至 4×10 个，包括 1×2 个，1×3 个，2×3 个，2×5 个，3×5 个，3×8 个，3×10 个，4×8 个，等等。腔室 2 的相邻两个腔室中心的间距为 4.5mm 的倍数，与实验室常规使用的 384 孔板或 96 孔板相一致，可配合多枪头加样枪的枪头间距(9mm)，还与自动点样机器人的点样针间距(4.5mm 的倍数)一致，便于加样、点样操作。

如图 1、图 8 和图 9 所示，腔室 2 的相邻两个腔室中心的间距为 4.5mm。

如图 5、图 6 和图 7 所示，腔室 2 的相邻两个腔室中心的间距为 9mm。

如图 4 所示，腔室 2 的相邻两个腔室中心的间距为 18mm。腔室 2 的相邻两个腔室中心的间距也可以为 13.5mm。

例如：腔室边长为 3mm，两个相邻腔室的中心间距则为 4.5mm。腔室边长为 6mm，两个相邻腔室的中心间距则为 9mm。腔室边长为 9mm，两个相邻腔室的中心间距则为 13.5mm。腔室边长为 12mm，两个相邻腔室的中心间距则为 18mm。

每个腔室 2 内布有微阵列探针点，可同时完成多种反应，即在每个腔室 2 中同时完成多种反应。在实验中节约了样品，最少时只需 5μl 即可完成分析检测。

本发明的多样品微阵列生物芯片用于蛋白质杂交、核酸（脱氧核糖核酸 DNA 或核糖核酸 RNA）杂交、脱氧核糖核酸(DNA)扩增。整个芯片可以承受高达 97℃温度，承受－40℃的低温，可以耐受紫外处理，经受水流清洗。

参看图 10 和图 11，在本发明的一个多样品微阵列生物芯片上同时检测两个样品，左侧腔室显示该样品反应阳性，右侧腔室则显示另一样品反应阴性。

本发明的多样品微阵列生物芯片制作和使用过程如下：

1、使用镊子将覆盖于单面胶材上的纸轻轻剥离，将单面胶材带有粘性的一面仔细覆盖粘贴于载玻片上，粘贴时注意保持载玻片洁净干燥，并注意不要让气泡进入粘贴面。把载玻片翻过来， 置于洁净平面，用镊子背敲打载玻片，使之

粘贴更紧密。

2、根据所需检测的项目确定每一个腔室中的点阵和方阵，并根据所设计的点阵和方阵使用点样机器人进行点样。

3、根据反应需要和反应条件，在每个腔室中加入不同样品和反应试剂。

4、仔细地将聚酯薄膜带有粘性的一面，从单面胶材的一侧开始整齐地粘贴于单面胶材的表面，即可进行孵育等反应。

5、待反应结束后，将聚酯薄膜撕下，可以进行清洗；如需再加样，则在加样后重新粘贴一层聚酯薄膜，继续进行反应。

6、当所有反应完毕后，最后将单面胶材撕除，即可用芯片扫描仪观察结果。在撕除单面胶材时可以用刀片轻轻地切割，这样操作会更方便。

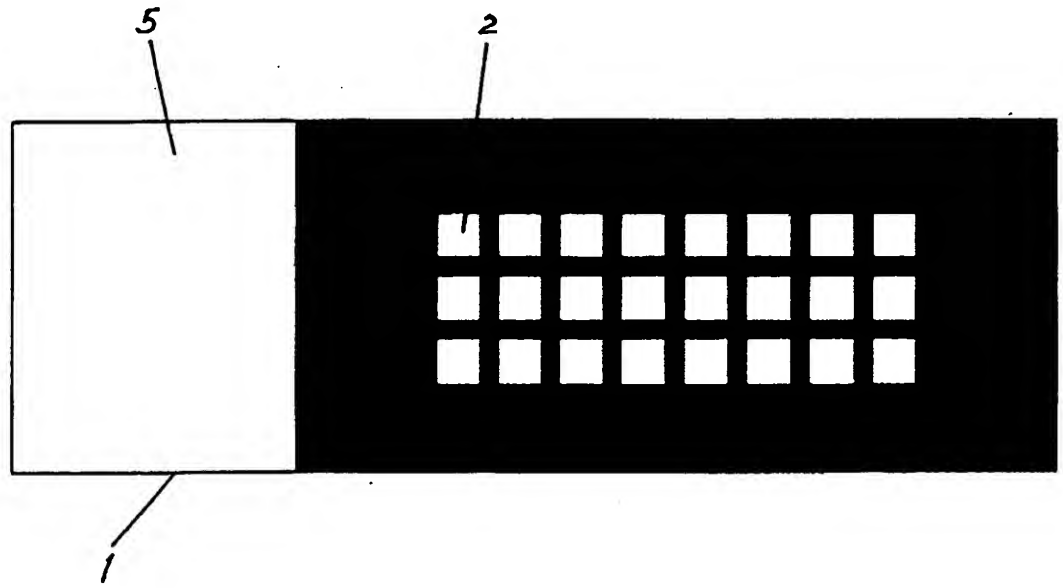

图 1

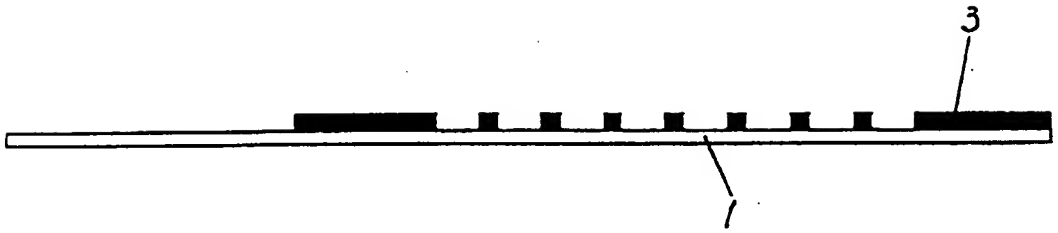

图 2

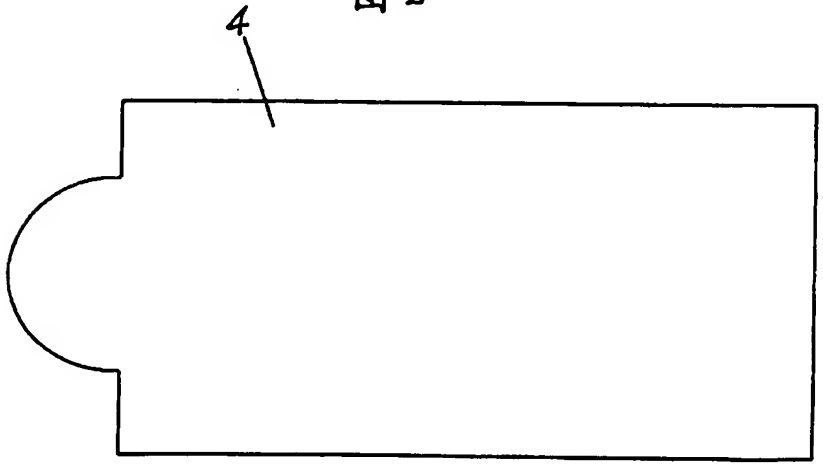

图 3

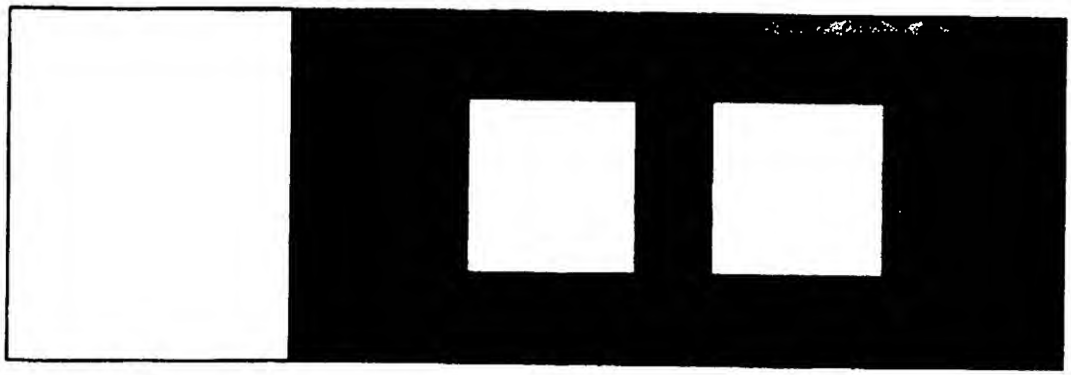

图 4

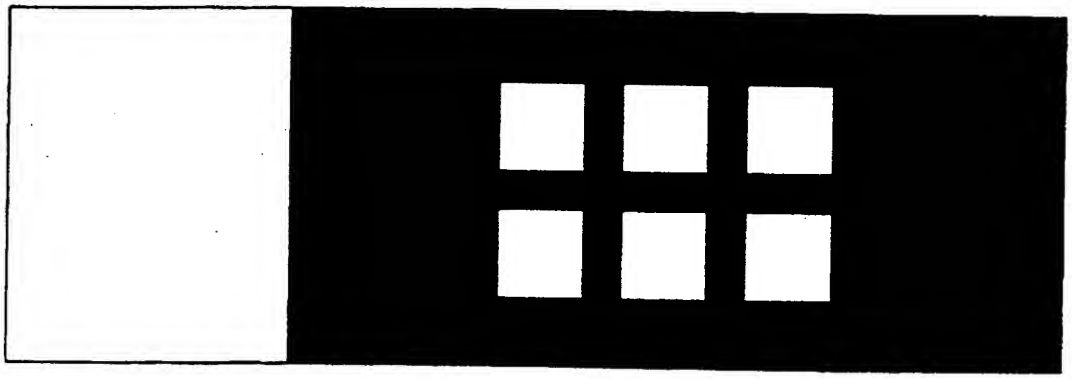

图 5

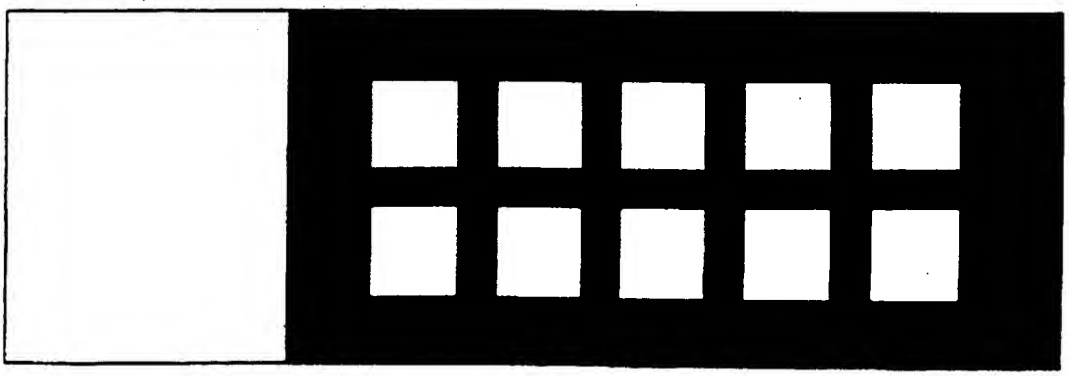

图 6

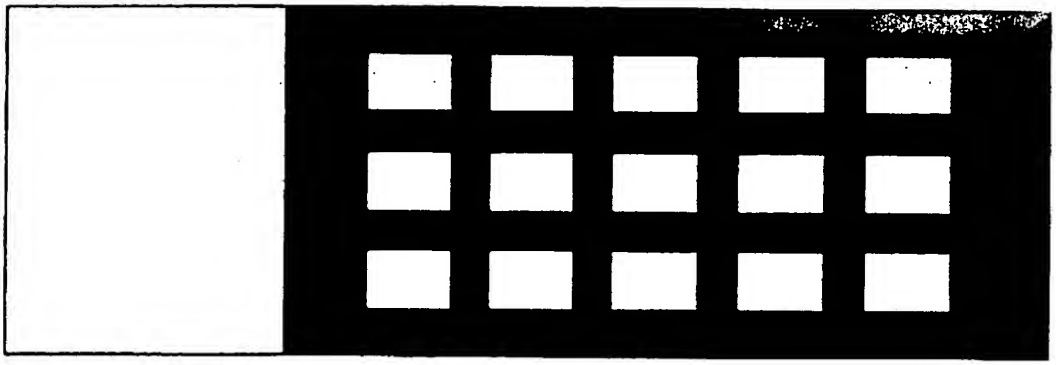

图 7

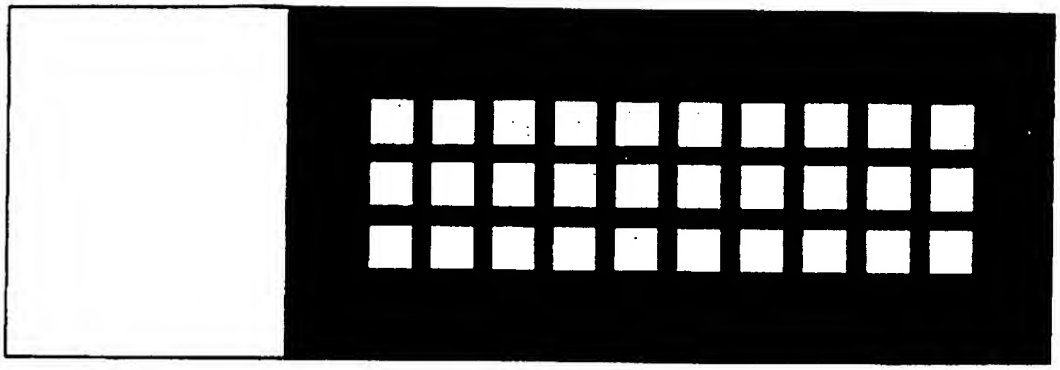

图 8

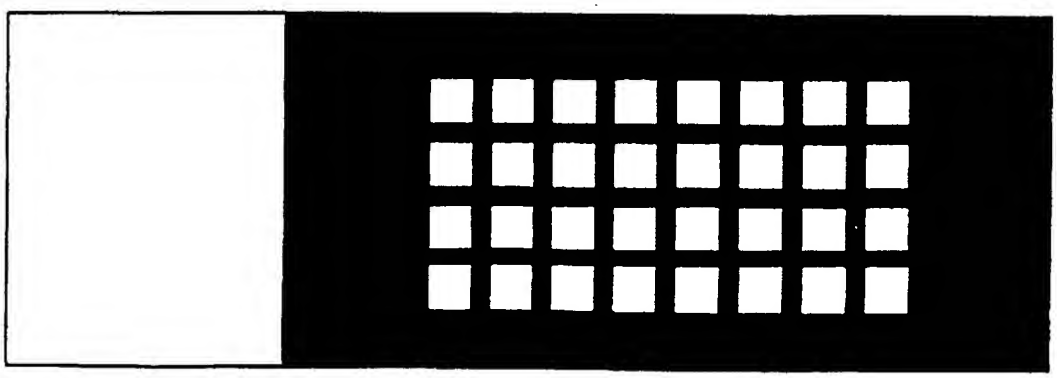

图 9

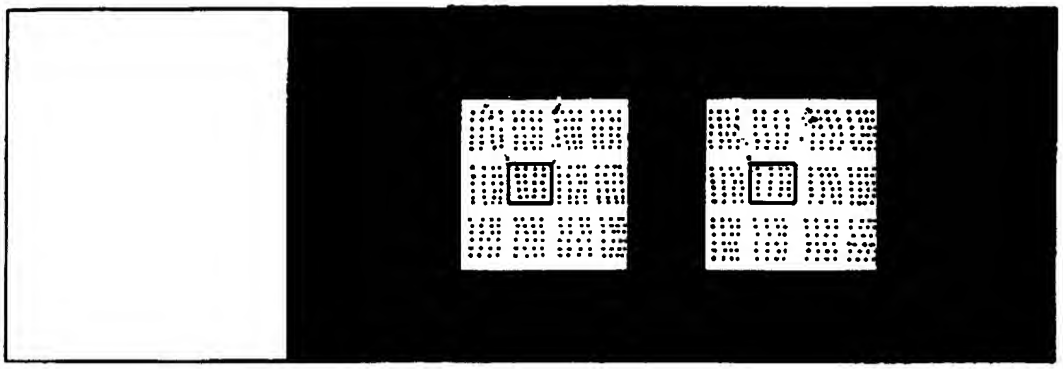

图 10

图 11

多样品微阵列生物芯片

(Chinese patent application number 01112783.x)

A kind of microarray biochip for multi-sample assay

Claims

1.A kind of microarray biochip for multi-sample assay, comprises of a glass solid support, on the surface of said glass solid support there are separate chambers which cannot leak each other and are formed by a single film of adhesive tape with lattices stick on the surface of the support, and a film of polyester is stick on the surface of the adhesive tape to close the chambers; there are probe dots in every chamber to perform simultaneity multi-reaction.

2. The biochip in claim 1 is applied to protein hybridization, nucleic acid hybridization and DNA amplification.

3. The biochip in claim 1or 2, wherein the length of the side of the chamber is the multiple of 3 mm.

4. The biochip in claim 1or 2, the space between the centers of two close chambers is the multiple of 4.5 mm.

5. The biochip of claim 3, wherein the space between the centers of two close chambers is 4.5 mm.

6. The biochip of claim 3, wherein the space between the centers of two close chambers is 9 mm.

7. The biochip of claim 3, wherein the space between the centers of two close chambers is 13.5 mm.

8. The biochip of claim 3, wherein the space between the centers of two close chambers is 18 mm.

9. The biochip of claim 1or 2, the number of said chambers are 1×2 to 4×10.

Description

This invention refers to the area of life science, especially a kind of microarray biochip for multi-sample assay with multiple samples arrayed orderly on its solid support.

The present microarray biochip, whose solid support is the plane medium that doesn't be treated specially, e.g. take common glass slide or pyroxylin film. This biochip in one reaction can only react with one non-solid bio-sample. When lots of samples need the

same testing, every sample had to be tested respectively, it is time wasted and affecting the parallelity and comparison mutual all the samples if there is any different between the treatment conditions (e.g. temperature and reaction time), so the aims of parallel experiments of multiple samples and items cannot be achieved. The reaction using this biochip, the non-solid bio-sample had to cover a larger area, large quantitative of sample is needed, and the flowing and vaporizing of the sample during the course of reaction, so they can make error.

The aim of this invention is providing a kind of microarray biochip for multi-sample assay, which can perform the reactions of test、analysis、identification and hybridization, and multiple samples and items parallel experiments.

The technical scheme of this invention as follows:
A kind of microarray biochip for multi-sample assay, comprises of a glass solid support, on the surface of said glass solid support there are separate chambers which cannot leak each other and are formed by a single film of adhesive tape with lattices stick on the surface of the support, and a film of polyester is stick on the surface of the adhesive tape to close the chambers; there are probe dots in every chamber to perform simultaneity multi-reaction.
The biochip is applied to protein hybridization, nucleic acid hybridization and DNA amplification.

The side length of the chamber of the biochip is the multiple of 3 mm.

The space between the centers of two close chambers of the biochip is the multiple of 4.5 mm.

The microarray biochip for multi-sample assay in this invention is a experiment device for RNA, DNA, protein and polypeptide, its surface of said glass solid support has bio-sample array, it can perform the reactions of test、analysis、identification and hybridization. Its size is small, it is easy for storage, operation and produce, the productive cost is low, it can be scanned with common biochip scanner.
This invention is a new reactive mode and can be applied to protein hybridization, DNA hybridization and DNA amplification. Since there are separate chambers on the glass slide, and each of the chambers is fixed microarray and spotted probes, such as DNA, RNA, protein or polypeptide, so it can reach the aim of multiple samples and items parallel experiments. If it is used in clinical, it can do synchronously the diagnosises of multiple samples, multiple items and multiple diseases.
The following will give detailed description with the figures and examples.

Figure 1 shows the sketch map of the structure of the microarray biochip for multi-sample assay, the number of the chambers are 3×8.

Figure 2 is an looking up sketch map of figure 1.
Figure 3 shows the sketch map of the structure of polyester adhering the biochip of figure 1.
Figure 4 shows the sketch map of the chambers of a biochip are 1×2.
Figure 5 shows the sketch map of the chambers of a biochip are 2×3.
Figure 6 shows the sketch map of the chambers of a biochip are2×5
Figure 7 shows the sketch map of the chambers of a biochip are 3×5.
Figure 8 shows the sketch map of the chambers of a biochip are 3×10.
Figure 9 shows the sketch map of the chambers of a biochip are 4×8.
Figure 10 shows the sketch map of testing two samples simultaneously on the biochip of this invention.
Figure 11 is the enlarged figure of the two samples of figure 10.

A kind of microarray biochip for multi-sample assay, comprises of a glass solid support, on the surface of said glass solid support there are separate chambers
Referring to figure 1 and figure 3: this invention is a kind of biochip comprises of a glass solid support. Glass slide(1) is the same as the standard glass slide of microscope with size of 25×75×0.96mm.

On the surface of said glass slide(1) there are separate chambers(2) which cannot leak each other, the chambers are formed by a single film of adhesive tape(3) with lattices stick on the surface of the support, and a film of polyester(4) is stick on the surface of the adhesive tape to close the chambers. The blank region without adhesive tape(3) and the film of polyester(4) is for label(5).

The size of adhesive tape(3) with lattices are 50mm×25mm suit to the size of the glass slide. The thickness of adhesive tape is at least 1mm(1mm-3mm), its one side is stickiness which can stick to the surface of glass slide(1) that is separated into small chambers(2) which cannot leak each other. The adhesive tape(3) is not dissolved when immersed in water and it can be washed, it is not transfiguration and leakage when used on the glass slide. After the experiment, take off the adhesive tape(3) and the biochip can be tested and analyzed by biochip scanner.
The film of polyester(4) is sticked on the surface of adhesive tape(3) with lattices. The one side of the film of polyester(4) is stickiness which can stick to the non-adhesive surface of adhesive tape(3) to seal up the chambers(2). The stickiness of the film of polyester(4) is lower than that of the film of polyester(4), which can pull apart easily and the stick between adhesive tape(3) and glass slide is not affected. After adhesive tape(3) adhered on glass slide(1) is covered with the film of polyester(4), separative closed chambers(2) are formed. There is no cross-contamination and the vapor is lower, so the accuracy of reaction is enhanced.

The side length of every chamber(2) is the multiple of 3mm. The number of the chambers are 1×2 to 4×10, comprising 1×2, 1×3, 2×3, 2×5, 3×5, 3×8, 3×10, 4×8, etc. The space between the centers of two close chambers is the multiple of 4.5mm corresponding to the microplate with holes of 384 or 96 used in laboratory. So the biochip is easily adapted the multichannel pippet with the space of 9mm between two tips, and also is adapted the autospoter with the space of the multiple of 4.5mm between spotting needles, easily for the operations of sample adding and spotting.

In figure 1, 8 and 9, the space between the centers of two close chambers(2) is 4.5mm.
In figure 5, 6 and 7, the space between the centers of two close chambers(2) is 9mm.
In figure 4, the space between the centers of two close chambers(2) is 18mm.
The space between the centers of two close chambers(2) can also be 13.5mm.

For example, the length of chamber is 3mm, so the space between the centers of two close chambers is 4.5mm; the side length of the chamber is 6mm, so the space between the centers of two close chambers is 9mm. The side length of the chamber is 9mm, so the space between the centers of two close chambers is 13.5mm. The side length of chamber is 12mm, the space between the centers of two close chambers is 18mm.

There are microarray probes in every chamber(2), which can react simultaneously, namely every chamber can react simultaneously. In this experiment, the sample is saving, the minimum only 5ul sample can fulfill the test.

Said biochip is applied to protein hybridization, nucleic acid(DNA or RNA) hybridization and DNA amplification. The whole biochip can endure the temperature to even 97℃ and low to -40℃, and can also endure ultraviolet and wash.

Referring to figure 10 and figure 11: on the said biochip, two samples are tested simultaneously. The left chamber shows positive reaction of a sample and the right shows negative reaction of another sample.

The course of formation and usage of said biochip as following:
1. The paper covered on the adhesive tape is rip off slightly by nipper, put the adhesive tape carefully with sticky side over the glass slide, when sticking the glass slide keeps clean and dry, and not let air bubble come into sticking plane. The glass is turn over, put on a clean plane and pressed to stick close.
2. The spots and the arrays in every chamber are conformed according to the item to be tested, and the samples are spotted according to the spots and the arrays designed.
3. Each of the chambers is added different sample and reactive agents according to the reactive aim and condition.

4. The film of polyester is sticked carefully on the surface of the adhesive tape, the reactions such as incubation of the biochip can be done.
5. After reaction the film of polyester is taken off and the biochip can be washed; if more sample added step is need, repeat step 3 and step 4.
6. After all the reactions finish, the adhesive tape is taken off finally, and the result of the test is processed by biochip scanner. When the adhesive tape is taken off, you can incise carefully using blade. It is more suitable.